UNIVERSITY OF WASHINGTON
School of Medicine
Department of Preventive Medicine
Seattle, Washington 98105

October 30, 1964

Office of Naval Research
Medicine and Dentistry Branch (Code 444)
Washington 25, D. C.

Re: Nonr(G)-00034-64
Project NR 105-322

Dear Sir:

Enclosed are the required copies of the Annual Scientific Report for the above contract as requested by Mr. Walter D. Smith, Resident Representative, Office of Naval Research, University of Washington.

Sincerely yours,

J. Thomas Grayston, M. D.
Professor and Chairman

JTG:lf

cc: Armed Services Technical Information Agency
Documents Service Center
Arlington Hall Station
Arlington 12, Virginia

Office of Naval Research Branch Office
230 N. Michigan Avenue
Chicago, 1, Illinois

Office of Naval Research Branch Office
207 West 24th Street
New York 11, New York

Office of Naval Research Branch Office
1030 East Green Street
Pasadena 1, California

Director, Naval Research Laboratory
Attn: Technical Information Officer
Washington 25, D. C.

Office of Technical Services
Department of Commerce
Washington 25, D. C.

U.S. Navy Office of Naval Research
Branch Office, Box 39
Navy No. 100, Fleet Post Office
New York, New York

Mr. Walter D. Smith
Resident Representative
University of Washington
Office of Naval Research
Seattle, Washington

TRACHOMA VIRUS RESEARCH

H. M. Jenkin
Department of Preventive Medicine
University of Washington

and

U. S. Naval Medical Research Unit No. 2
Taipei, Taiwan, ROC

PROJECT NR 105-322 CONTRACT Nonr (G)-00034-64

OBJECTIVES

To study the dynamics of growth, immunological and chemical characterization of PLT agents using cell culture, biochemical, immunological and radioisotope tools. This work was not confined to the PLT agents. PPLO and viruses such as influenza and arboviruses were also under investigation.

ABSTRACT

An abstract of results of continuous passage of trachoma in HeLa cells has been published (Jenkin and O'Donnell, 1963). Recently 47 and 53 cell culture passage of trachoma strains, TW3 and Bour, respectively, showed continued pathogenicity in monkey eyes. The lack of second cycles of growth of trachoma in the HeLa cell system seems associated with interferon production. In this laboratory trachoma does not produce a CPE in HeLa cells permitting the use of Chikungunya, a group A arbovirus which does cause a CPE, as a challenge organism for detection of interferon.

Lipid analysis of host (chick allantoic fluid - "L" cells) and purified PLT organisms (6BC and meningopneumonitis virus) using thin layer and gas chromatography techniques showed quantitative and qualitative differences in the phospholipid fractions between the host and parasite as well as between parasite strains. Gas chromatography and thin layer chromatography analysis of fatty acids of phospholipid species of different strains of PLT is under investigation. ( )

Determination of necessary levels of C^{14} serine for tracer studies of lipid metabolism of PLT agents in cell culture is under investigation. Using this C^{14} marker, phospholipid synthesis in normal host cells has been followed using column and thin layer chromatography methods for separation and detection of the labelled components. The label was readily traced to the phosphotydl ethanolamine fractions.

Trachoma Virus Research

Preparation of cell wall antigens originally cultivated in two host systems and preparation of antibodies to PLT agents and normal host antigens in several animal species has been in progress as a preliminary for serological differentiation of strains of the PLT group and following persistence of antibody response.

Arbovirus Studies

An arbovirus serum survey of an Indonesian population using cell culture neutralization techniques in a hamster embryonic cell line established here has shown that a Chikungunya-like, Japanese B encephalitis - West Nile-like, and Bunyamwera-like viruses were in the Djakarta area. Identification of prototype dengue virus and dengue virus antibody levels has been accomplished recently using interferon (SEATO laboratory technique) and plaque reduction tests. This virus can now be screened to finish the serum survey.

Eaton PPLO

An Eaton PPLO (M. pneumoniae) epidemic which occurred between the middle of June and the end of July, 1964 among American military personnel in Taipei was studied. Eighty-seven percent of 21 cases showed significant CF antibody levels against a new Eaton PPLO lipid antigen, (antigen preparation made by George Kenny). One isolate was obtained and identified by morphological, cultural and hemolytic properties. Serological identity is now in progress. An abstract by Jenkin and Picken, 1964 (to be published), gives further details of this study.

Influenza Surveillance

An epidemic of influenza A_2 occurring in January-February, 1964 was investigated. Eight isolates out of 86 selected cases were obtained which were antigenically similar to A_2/Japan/305/57 and recent A_2 Taiwan influenza strains. Seventy percent of paired serums $(48)^2$ from Taipei Provincial Children's Hospital showed at least four-fold rises in hemagglutination-inhibition (HI) antibody titers. Adult serums obtained from NAMRU-2 personnel showed a 38% (162 paired serums) four-fold rise in HI titer. A long term study of persistance of influenza antibodies after active infection showed that HI antibody levels dropped markedly between 180-220 days after onset of illness. Those antibody titers which were less than 1:512 25 to 35 days after onset essentially disappeared, whereas HI titers at 1:512 or greater at that time still showed circulating antibody at 180 days. These studies will be continued.

TRACHOMA VIRUS RESEARCH

H. M. Jenkin
Department of Preventive Medicine
University of Washington

and

U. S. Naval Medical Research Unit No. 2
Taipei, Taiwan, ROC

PROJECT NR 105-322 CONTRACT Nonr (G)-00034-64

OBJECTIVES

To study the dynamics of growth, immunological and chemical characterization of PLT agents using cell culture, biochemical, immunological and radioisotope tools. This work was not confined to the PLT agents. PPLO and viruses such as influenza and arboviruses were also under investigation.

ABSTRACT

An abstract of results of continuous passage of trachoma in HeLa cells has been published (Jenkin and O'Donnell, 1963). Recently 47 and 53 cell culture passage of trachoma strains, TW3 and Bour, respectively, showed continued pathogenicity in monkey eyes. The lack of second cycles of growth of trachoma in the HeLa cell system seems associated with interferon production. In this laboratory trachoma does not produce a CPE in HeLa cells permitting the use of Chikungunya, a group A arbovirus which does cause a CPE, as a challenge organism for detection of interferon.

Lipid analysis of host (chick allantoic fluid - "L" cells) and purified PLT organisms (6BC and meningopneumonitis virus) using thin layer and gas chromatography techniques showed quantitative and qualitative differences in the phospholipid fractions between the host and parasite as well as between parasite strains. Gas chromatography and thin layer chromatography analysis of fatty acids of phospholipid species of different strains of PLT is under investigation.

Determination of necessary levels of C^{14} serine for tracer studies of lipid metabolism of PLT agents in cell culture is under investigation. Using this C^{14} marker, phospholipid synthesis in normal host cells has been followed using column and thin layer chromatography methods for separation and detection of the labelled components. The label was readily traced to the phosphotydl ethanolamine fractions.

Trachoma Virus Research

Preparation of cell wall antigens originally cultivated in two host systems and preparation of antibodies to PLT agents and normal host antigens in several animal species has been in progress as a preliminary for serological differentiation of strains of the PLT group and following persistence of antibody response.

Arbovirus Studies

An arbovirus serum survey of an Indonesian population using cell culture neutralization techniques in a hamster embryonic cell line established here has shown that a Chikungunya-like, Japanese B encephalitis - West Nile-like, and Bunyamwera-like viruses were in the Djakarta area. Identification of prototype dengue virus and dengue virus antibody levels has been accomplished recently using interferon (SEATO laboratory technique) and plaque reduction tests. This virus can now be screened to finish the serum survey.

Eaton PPLO

An Eaton PPLO (M. pneumoniae) epidemic which occurred between the middle of June and the end of July, 1964 among American military personnel in Taipei was studied. Eighty-seven percent of 21 cases showed significant CF antibody levels against a new Eaton PPLO lipid antigen, (antigen preparation made by George Kenny). One isolate was obtained and identified by morphological, cultural and hemolytic properties. Serological identity is now in progress. An abstract by Jenkin and Picken, 1964 (to be published), gives further details of this study.

Influenza Surveillance

An epidemic of influenza A_2 occurring in January-February, 1964 was investigated. Eight isolates out of 86 selected cases were obtained which were antigenically similar to A_2/Japan/305/57 and recent A_2 Taiwan influenza strains. Seventy percent of paired serums $(48)^2$ from Taipei Provincial Children's Hospital showed at least four-fold rises in hemagglutination-inhibition (HI) antibody titers. Adult serums obtained from NAMRU-2 personnel showed a 38% (162 paired serums) four-fold rise in HI titer. A long term study of persistance of influenza antibodies after active infection showed that HI antibody levels dropped markedly between 180-220 days after onset of illness. Those antibody titers which were less than 1:512 25 to 35 days after onset essentially disappeared, whereas HI titers at 1:512 or greater at that time still showed circulating antibody at 180 days. These studies will be continued.

Trachoma Virus Research

Preparation of cell wall antigens originally cultivated in two host systems and preparation of antibodies to PLT agents and normal host antigens in several animal species has been in progress as a preliminary for serological differentiation of strains of the PLT group and following persistence of antibody response.

Arbovirus Studies

An arbovirus serum survey of an Indonesian population using cell culture neutralization techniques in a hamster embryonic cell line established here has shown that a Chikungunya-like, Japanese B encephalitis - West Nile-like, and Bunyamwera-like viruses were in the Djakarta area. Identification of prototype dengue virus and dengue virus antibody levels has been accomplished recently using interferon (SEATO laboratory technique) and plaque reduction tests. This virus can now be screened to finish the serum survey.

Eaton PPLO

An Eaton PPLO (M. pneumoniae) epidemic which occurred between the middle of June and the end of July, 1964 among American military personnel in Taipei was studied. Eighty-seven percent of 21 cases showed significant CF antibody levels against a new Eaton PPLO lipid antigen, (antigen preparation made by George Kenny). One isolate was obtained and identified by morphological, cultural and hemolytic properties. Serological identity is now in progress. An abstract by Jenkin and Picken, 1964 (to be published), gives further details of this study.

Influenza Surveillance

An epidemic of influenza A_2 occurring in January-February, 1964 was investigated. Eight isolates out of 86 selected cases were obtained which were antigenically similar to A_2/Japan/305/57 and recent A_2 Taiwan influenza strains. Seventy percent of paired serums $(48)^2$ from Taipei Provincial Children's Hospital showed at least four-fold rises in hemagglutination-inhibition (HI) antibody titers. Adult serums obtained from NAMRU-2 personnel showed a 38% (162 paired serums) four-fold rise in HI titer. A long term study of persistance of influenza antibodies after active infection showed that HI antibody levels dropped markedly between 180-220 days after onset of illness. Those antibody titers which were less than 1:512 25 to 35 days after onset essentially disappeared, whereas HI titers at 1:512 or greater at that time still showed circulating antibody at 180 days. These studies will be continued.